APA860Mu02 100μg
Active Epithelial Neutrophil Activating Peptide 78 (ENA78)
Organism Species: *Mus musculus (Mouse)*
Instruction manual

FOR RESEARCH USE ONLY
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

12th Edition (Revised in Aug, 2016)

[PROPERTIES]

Source: Prokaryotic expression.
Host: *E. coli*
Residues: Ala41~Gln132
Tags: N-terminal His-tag
Purity: >97%
Traits: Freeze-dried powder
Endotoxin Level: <1.0EU per 1μg (determined by the LAL method).
Buffer Formulation: PBS, pH7.4, containing 0.01% SKL, 5% Trehalose.
Original Concentration: 200μg/mL
Applications: Cell culture; Activity Assays.
(May be suitable for use in other assays to be determined by the end user.)
Predicted isoelectric point: 9.5
Predicted Molecular Mass: 13.6kDa
Accurate Molecular Mass: 14kDa as determined by SDS-PAGE reducing conditions.

[USAGE]

Reconstitute in 10mM PBS (pH7.4) to a concentration of 0.1-1.0 mg/mL. Do not vortex.

[STORAGE AND STABILITY]

Storage: Avoid repeated freeze/thaw cycles.

Store at 2-8°C for one month.

Aliquot and store at -80°C for 12 months.

Stability Test: The thermal stability is described by the loss rate. The loss rate was determined by accelerated thermal degradation test, that is, incubate the protein at 37°C for 48h, and no obvious degradation and precipitation were observed. The loss rate is less than 5% within the expiration date under appropriate storage condition.

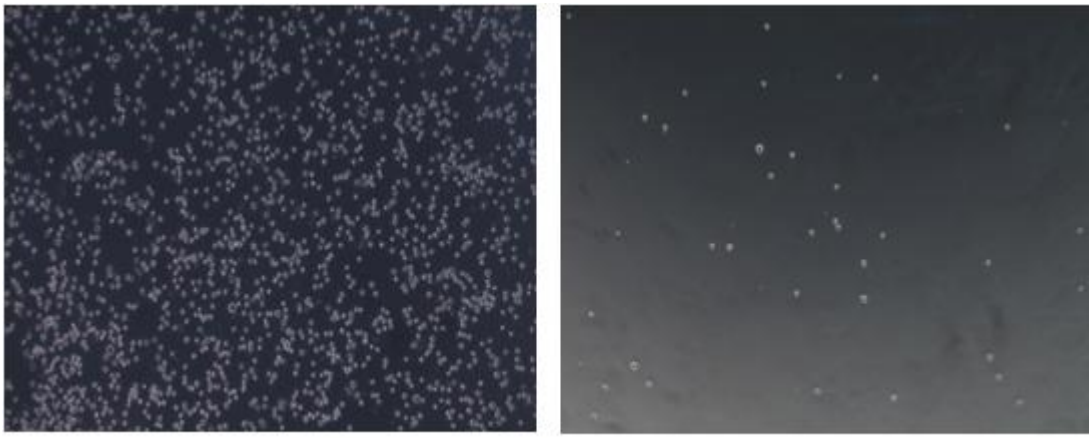
[SEQUENCE]

APSSVIAATE
LRCVCLTVTP KINPKLIANL EVIPAGPQCP TVEVIAKLKN QKEVCLDPEA
PVIKKIIQKI LGSDKKKAKR NALAVERTAS VQ

[ACTIVITY]

Epithelial Neutrophil Activating Peptide 78 (ENA-78) is a member of the CXC subfamily of chemokines. Full-length ENA-78 is 114 amino acids (aa) in length with a predicted molecular weight of 12 kDa. ENA-78 is expressed by many immune cells, such as macrophages, eosinophils, as well as non-immune cells including mesothelial cells, and fibroblasts. Soluble ENA-78 potently chemoattracts T cells and monocytes, while the cell-bound chemokine promotes strong adhesion of leukocytes to activated endothelial cells, where it is primarily expressed. Thus, chemotaxis assay used 24-well microchemotaxis system was undertaken to detect the chemotactic effect of ENA-78 on THP-1 the human monocytic cell line. Briefly, THP-1 cells were seeded into the upper chambers (150 ul cell suspension,10^6 cells/ml in RPMI 1640 with FBS free) and different concentrations of recombinant mouse ENA-78 diluted with serum free RPMI 1640 was added in lower chamber with a polycarbonate filter (8 um pore size) used to separate the two compartments.

After incubation at 37 ℃ with 5% CO_2 for 1h, the filter was removed, then cells in low chamber were observed by inverted microscope at low magnification (×10) and the number of migrated cells were counted using Fluorescence Activating Cell Sorter. Result shows ENA-78 is able to induce migration of THP-1 cells. The migrated THP-1 cells in low chamber at low magnification (×10) were shown in Figure 1. Statistical results of FACS were shown in Figure 2. The optimum chemotaxis of ENA-78 occurs at 1 ng/ml.



A B

Figure 1. The chemotactic effect of ENA-78 on THP-1 cells

(A) Cells in lower chamber after incubation at 37 ℃for 1h in which THP-1 cells were seeded into the upper chambers and 1 ng/ml recombinant mouse ENA-78 was added in lower chamber.

(B) Cells in lower chamber after incubation at 37 ℃for 1h in which THP-1 cells were seeded into the upper chambers and serum free RPMI 1640 without recombinant mouse ENA-78 was added in lower chamber.

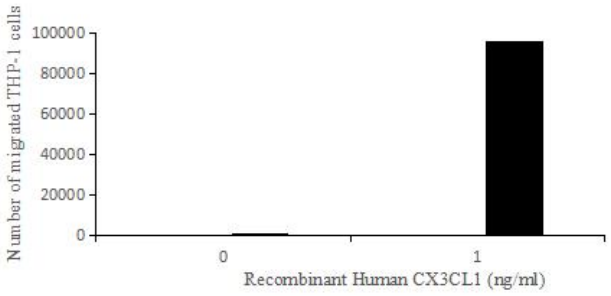


Figure 2. The chemotactic effect of ENA-78 on THP-1 cells

[IDENTIFICATION]

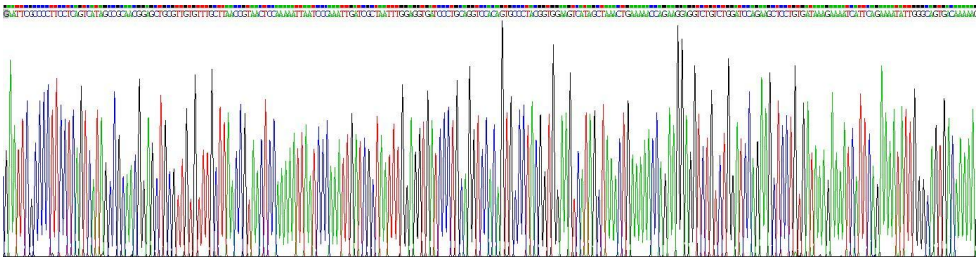


Figure 3. Gene Sequencing (extract)

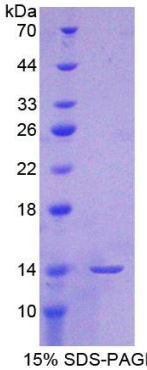


Figure 4. SDS-PAGE

Sample: Active recombinant ENA78, Mouse

[IMPORTANT NOTE]

The kit is designed for research use only, we will not be responsible for any issue if the kit was used in clinical diagnostic or any other procedures.